# ETC RAPID DETECTION OF MICROBIAL CONTAMINATION OF WATER ACTION TEAM PROGRESS REPORT

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**Environmental Problem:** Waterborne pathogens continue to contaminate drinking water supplies and cause waterborne disease outbreaks (WBDO) despite current regulations designed to prevent and control their spread. Annually, the CDC estimates that pathogen infected drinking water results in about a million new cases of illness and about a thousand deaths.

EPA currently regulates two indicators of microbiological drinking water quality: total coliform and turbidity. Challenges of using indicator organisms for monitoring water quality are: (1) Poor correlation between indicators and the presences of pathogens since there is a great diversity of microbial pathogens; and (2) Long delay in obtaining results thereby causing a time lag between the occurrence of the contamination event and its detection to be able to safe guard consumer health.

Therefore, "rapid" or "near real-time" quantitative analytical methods are needed that can specifically detect a broad array of microorganisms.

#### **Technology Challenge:**

# 1. Development of Molecular Detection Technology for Monitoring Water

Molecular detection technologies have the ability to detect multiple pathogens in a single analysis, to make highly specific identifications, and to detect very low numbers of target organisms rapidly. Although a considerable amount of work has been done to develop rapid, sensitive, and quantitative molecular methods, several challenges remain that must be resolved before EPA will approve these methods for routine monitoring of environmental waters. An important one is, prior to water analysis, steps involved in concentrating a sample must be developed to reliably give the needed sensitivity to detect the low number of pathogens that may be present in water samples.

### 2. Comparison of Molecular Detection Technologies to Existing Methods

Existing methods are based on the detection of culturable indicator organisms. Results from molecular methods must be related to the existing methods for detection of viable organisms. Additionally molecular methods must have equal or greater detection sensitivity to indicator bacteria methods (1 per 100 mL). Molecular methods must also be able to detect specific pathogens, a characteristic which existing methods lack, including non-culturable pathogens.

# 3. Acceptance of Molecular Methods

The water industry has used existing indicator bacteria methods for approximately 100 years. To gain acceptance, molecular methods must satisfactorily demonstrate that they provide useful data on pathogens that has the same or greater predictive value to that of methods for indicator organisms.

The mission of the action team will be to (1) identify innovative technologies that demonstrate the greatest potential for environmental application (2) identify research gaps in adapting molecular technologies for consideration by ORD's Research Program, including the STAR and SBIR Extramural Programs, and AWWARF (3) provide technical support for the expansion of methods and (4) quantify economic and environmental benefits for the use of these methods.

#### FY'06 Accomplishments:

- Redefined our mission to make it problem focused rather than being technology focused. Thus, rather than concentrating on the use of microarray and related technologies to monitor water, our mission has been restated to determining microbial contamination of water using rapid and sensitive molecular-based technologies.
- Office of Water has performed a literature review of microarray technologies. The work was completed on March 31, 2005 and is available on-line at epa.gov/safewater/ucmr/pdfs/summary\_workshop\_microarrays\_bibliography.pdf
- A workshop to solicit ideas, suggestions and recommendations on the feasibility
  of using DNA microarrays for high-throughput detection of waterborne pathogens
  was held on March 21-22, 2005, at Cincinnati by the Office of Water. The
  proceedings of the workshop are posted at
  <a href="https://www.epa.gov/safewater/ucmr/pdfs/summary\_workshop\_microarrays.pdf">workshop\_microarrays.pdf</a>
- Based on a need identified at the above workshop, a second workshop was
  organized jointly by OW and ORD to discuss a wide variety of techniques for
  detecting, capturing and concentrating waterborne pathogens from large volume
  of water, in April, 2006 at Cincinnati.
- As a consequence of the second workshop a memorandum of understanding was signed between EPA and Sandia Labs for pursuing research on Insulator based Dielectrophoresis (IDEP) technology for concentrating pathogens for water samples.

# **General accomplishments:**

- Monthly meetings
- Added three new members: from the American Water Works Research Foundation, University of Arizona, AR, and National Center for Environmental Research, ORD, EPA

#### FY'07 Plan:

- Organize a workshop with EPA and outside scientists to find research gaps in adapting molecular techniques for detecting waterborne pathogens in environmental samples.
- Compile a list of EPA funded projects that utilize molecular technologies to detect waterborne pathogens in environmental samples.
- Perform a literature review of each respective technology, namely, qPCR, microfluidics and lab-on-a-chip, which could be used for waterborne pathogen detection.
- Perform a product review of commercially available products related to each respective technology, namely qPCR, microarray, microfluidics and lab-on-achip, which could be used for waterborne pathogen detection.
- Perform market research of vendors selling the various consumables, reagents and instrumentation needed for the above mentioned technologies.